

CLAIMS

What is claimed is:

1. A chimeric protein comprising a functional domain and a carrier domain, wherein the functional domain comprises an amino acid sequence which binds a selected substance and the carrier domain comprises an amino acid sequence which binds a mammalian cell surface receptor other than the mammalian cell surface receptor which normally binds the selected substance in the mammal.
- 5 10 2. The chimeric protein of Claim 1, wherein the mammalian cell surface receptor is a human cell surface receptor.
3. The chimeric protein of Claim 2, wherein the human cell surface receptor is selected from the group consisting of: human low density lipoprotein receptors, human transferrin receptors, human serum albumin receptors, human asialoglycoprotein receptors, human adenovirus receptors, human retrovirus receptors, human CD4, human lipoprotein (a) receptors, human immunoglobulin Fc receptor, human α -fetoprotein receptors, human LDLR-like protein (LRP) receptors, human acetylated LDL receptors, human mannose receptors and human mannose-6-phosphate receptors.
- 15 20 25 4. The chimeric protein of Claim 2, wherein the selected substance is LDL cholesterol and the functional domain comprises the amino acid sequence of the ligand-binding domain of the human low density lipoprotein receptor.

5. The chimeric protein of Claim 4, wherein the carrier domain comprises an amino acid sequence which binds a human cell surface receptor other than the low density lipoprotein receptor.
- 5 6. The chimeric protein of Claim 5, wherein the human cell surface receptor is the human transferrin receptor.
7. DNA encoding a chimeric protein, the chimeric protein comprising a functional domain and a carrier domain, wherein the functional domain comprises an amino acid sequence which binds a selected substance and the carrier domain comprises an amino acid sequence which binds a mammalian cell surface receptor other than the mammalian cell surface receptor which normally binds the selected substance in the mammal.
- 10 15 8. DNA of Claim 7 which encodes a chimeric protein wherein the mammalian cell surface receptor is a human cell surface receptor.
9. DNA of Claim 8, wherein the human cell surface receptor is selected from the group consisting of: human low density lipoprotein receptors, human transferrin receptors, human serum albumin receptors, human asialoglycoprotein receptors, human adenovirus receptors, human retrovirus receptors, human CD4, human lipoprotein (a) receptors, human immunoglobulin Fc receptors, human α -fetoprotein receptors, human LDLR-like protein (LRP) receptors, human acetylated LDL receptor, human mannose receptors and human mannose-6-phosphate receptors.
- 20 25 10. DNA encoding a chimeric protein comprising a functional

domain and a carrier domain, wherein the functional domain comprises the amino acid sequence of the ligand-binding domain of the human low density lipoprotein receptor and the carrier domain comprises an amino acid sequence which binds the human transferrin receptor.

5 11. An expression plasmid comprising:

a) a promoter;
10 b) DNA encoding a chimeric protein comprising a functional domain and a carrier domain, wherein the functional domain comprises an amino acid sequence which binds a selected substance and the carrier domain comprises an amino acid sequence which binds a mammalian cell surface receptor other than the mammalian cell surface receptor which normally binds the selected substance in the mammal,

15 wherein expression of the DNA encoding the chimeric protein is under the control of the promoter.

20 12. A genetically modified mammalian cell which expresses DNA encoding a chimeric protein, wherein the chimeric protein comprises a functional domain and a carrier domain, wherein the functional domain comprises an amino acid sequence which binds a selected substance and the carrier domain comprises an amino acid sequence which binds a mammalian cell surface receptor other than the mammalian cell surface receptor which normally binds the selected substance in the mammal.

25 13. The genetically modified mammalian cell of Claim 12, wherein the mammalian cell surface receptor is a human cell surface receptor.

14. The genetically modified mammalian cell of Claim 13, wherein the selected substance is LDL cholesterol and the human cell surface receptor is the human transferrin receptor.

5 15. A genetically modified mammalian cell of Claim 14, wherein the functional domain comprises the amino acid sequence of the ligand-binding domain of the human low density lipoprotein receptor.

10 16. A genetically modified mammalian cell of Claim 13, wherein the human cell surface receptor is selected from the group consisting of: human low density lipoprotein receptors, human transferrin receptors, human serum albumin receptors, human asialoglycoprotein receptors, human adenovirus receptors, human retrovirus receptors, human CD4, human lipoprotein (a) receptors, human immunoglobulin Fc receptors, human α -fetoprotein receptors, human LDLR-like protein (LRP) receptors, human acetylated LDL receptors, human mannose receptors and human mannose-6-phosphate receptors.

15 20 17. The genetically modified mammalian cell of Claim 12, wherein the mammalian cell is selected from the group consisting of: human cells, porcine cells, hamster cells, bovine cells, canine cells, mouse cells, rat cells, monkey cells, feline cells, rabbit cells, sheep cells and chimpanzee cells.

25 30 18. The genetically modified human cell of Claim 17, wherein the human cell is selected from the group consisting of: human fibroblasts, human keratinocytes, human epithelial cells, human ovary cells, human endothelial cells, human glial cells, human neural

cells, formed elements of the blood, human muscle cells, human hepatocytes and precursors of these human cell types.

19. The genetically modified mammalian cell of Claim 17,
5 wherein the mammalian cells are Chinese hamster ovary cells.
20. The genetically modified mammalian cell of Claim 17,
wherein the cell is an immortalized cell.
21. The genetically modified human cell of Claim 18,
10 wherein the cell is a primary cell or a secondary cell.
22. The genetically modified mammalian cell of Claim 12,
wherein the DNA encodes a chimeric protein in which the
functional domain comprises the amino acid sequence of
the ligand-binding domain of the human low density
15 lipoprotein receptor and the carrier domain comprises
an amino acid sequence of human transferrin which binds
the human transferrin receptor.
23. The genetically modified mammalian cell of Claim 17,
wherein the DNA encodes a chimeric protein in which the
functional domain comprises the amino acid sequence of
the ligand-binding domain of the human low density
lipoprotein receptor and the carrier domain comprises
an amino acid sequence of human transferrin which binds
20 the human transferrin receptor.
- 25 24. The genetically modified mammalian cell of Claim 18,
wherein the DNA encodes a chimeric protein in which the
functional domain comprises the amino acid sequence of
the ligand-binding domain of the human low density
lipoprotein receptor and the carrier domain comprises

an amino acid sequence of human transferrin which binds the human transferrin receptor.

25. The genetically modified mammalian cell of Claim 19, wherein the DNA encodes a chimeric protein in which the functional domain comprises the amino acid sequence of the ligand-binding domain of the human low density lipoprotein receptor and the carrier domain comprises an amino acid sequence of human transferrin which binds the human transferrin receptor.

10 26. The genetically modified mammalian cell of Claim 20, wherein the DNA encodes a chimeric protein in which the functional domain comprises the amino acid sequence of the ligand-binding domain of the human low density lipoprotein receptor and the carrier domain comprises an amino acid sequence of human transferrin which binds the human transferrin receptor.

15 27. The genetically modified mammalian cell of Claim 21, wherein the DNA encodes a chimeric protein in which the functional domain comprises the amino acid sequence of the ligand-binding domain of the human low density lipoprotein receptor and the carrier domain comprises an amino acid sequence of human transferrin which binds the human transferrin receptor.

20 28. A method of transporting low density lipoprotein into a cell, comprising combining low density lipoprotein with:

25 a) a chimeric protein which comprises a functional, domain and a carrier domain, wherein the functional domain comprises the amino acid sequence of the ligand-binding domain of human low density lipoprotein receptor and the carrier

domain comprises an amino acid sequence which binds a human cell surface receptor other than human low density lipoprotein receptor; and

5 b) a human cell bearing a surface receptor which binds the amino acid sequence of the carrier domain of the chimeric protein,

10 under conditions appropriate for binding of low density lipoprotein to the ligand binding domain of low density lipoprotein and for binding of the amino acid sequence of the carrier domain of the chimeric protein with the cell surface receptor on the human cell, whereby a complex of the chimeric protein and low density lipoprotein is formed and the complex is transported into the cell by endocytosis.

15 29. The method of Claim 28, wherein the carrier domain binds a human cell surface receptor selected from the group consisting of: human low density lipoprotein receptors, human transferrin receptors, human serum albumin receptors, human asialoglycoprotein receptors,

20 human adenovirus receptors, human retrovirus receptors, human CD4, human lipoprotein (a) receptors, human immunoglobulin Fc receptor, human α -fetoprotein receptors, human LDLR-like protein (LRP) receptors, human acetylated LDL receptors, human mannose receptors and human mannose-6-phosphate receptors.

25 30. The method of Claim 28, wherein the carrier domain comprises an amino acid sequence of human transferrin which binds the cell surface receptor on the human cell.

30 31. A method of lowering the level of low density lipoprotein in the bloodstream of an individual,

comprising administering to the individual a chimeric protein comprising a functional domain and a carrier domain, wherein the functional domain comprises the amino acid sequence of the ligand-binding domain of human low density lipoprotein receptor and the carrier domain comprises an amino acid sequence which binds a human cell surface receptor other than human low density lipoprotein receptor, wherein low density lipoprotein in the bloodstream of the individual binds the ligand binding domain of low density lipoprotein in the chimeric protein and the carrier domain of the chimeric protein binds a human cell surface receptor, whereby the chimeric protein having low density lipoprotein bound thereto is transported into the cell bearing the cell surface receptor, thereby lowering the level of low density lipoprotein in the bloodstream of the individual.

32. The method of Claim 31, wherein the carrier domain binds a human cell surface receptor selected from the group consisting of: human low density lipoprotein receptors, human transferrin receptors, human serum albumin receptors, human asialoglycoprotein receptors, human adenovirus receptors, human retrovirus receptors, human CD4, human lipoprotein (a), human immunoglobulin Fc receptor, human a-fetoprotein receptors, human LDLR-like protein (LRP) receptors, human acetylated LDL receptors, human mannose receptors and human mannose-6-phosphate receptors.

33. The method of Claim 31, wherein the carrier domain comprises an amino acid sequence of human transferrin which binds to the human transferrin receptor.

34. A pharmaceutical composition comprising the chimeric protein of Claims 1, 2, 3, 4, 5, or 6.

35. A pharmaceutical composition comprising the genetically modified cell of Claims 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, or 27.

36. A method of introducing a selected substance into a mammalian cell, comprising contacting a mammalian cell to a chimeric protein which binds the selected substance at a functional domain, and which binds a cell surface receptor on the mammalian cell at a carrier domain, such that the complex of the cell surface receptor, the chimeric protein, and the selected substance forms and the complex is bound to the cell surface receptor and transported into the cell by receptor-mediated endocytosis.

37. The method of Claim 36, wherein the selected substance is selected from the group consisting of: acetylated low density lipoprotein, apolipoprotein E4, tumor necrosis factor α , transforming growth factor β , a cytokine, an immunoglobulin, a hormone, glucose, a bile salt, a glycolipid, and a glycosaminoglycan.

38. A method of introducing a functional protein domain into a mammalian cell, comprising contacting a mammalian cell with a chimeric protein which has a functional domain and which binds a cell surface receptor on the mammalian cell at a carrier domain, such that the complex of the cell surface receptor, the chimeric protein, and the selected substance forms and the complex is bound to the cell surface receptor and

transported into the cell by receptor-mediated endocytosis.

39. The method of Claim 38, wherein the functional protein domain comprises an amino acid sequence with enzymatic 5 activity.

40. The method of Claim 38, wherein the functional protein domain comprises an amino acid sequence which binds to a DNA or RNA sequence and increases or decreases gene expression in the mammalian cell.

10 41. The method of Claim 38, wherein the functional protein domain binds to a selected protein, carbohydrate, lipid, or glycolipid target in the mammalian cell.

42. The method of Claims 36, 37, 38, 39, 40, or 41, in which the mammalian cell is contacted with the chimeric 15 protein *in vivo*.

Add A1